

25 February 2005

The Commissioner of Patents
IP Australia
PO Box 200
Woden ACT 2606

Dear Madam

**International Patent Application No PCT/AU2004/001032
For 'Electrophoresis Membrane Support and Manifold Unit'**

In the name of Gradipore Limited

In response to the Written Opinion of the International Searching Authority dated 22 September 2004, the applicant offers the following comments and encloses substitute pages 1, 9, and 16 to 18 under Article 34(2).

The differences are as follows:

In the description, an error in the patent number of a US prior art document has been corrected on pages 1 and 9. The correct number is US 5039386. In the claims, amendment has been proposed to correct the antecedence problems noted in the Written Opinion in claims 1 and 17. Claim 15 has been amended to remove the text "any one of" from the preamble.

Marked-up copies of the description and claims are also provided for convenience to indicate the proposed amendments.

In response to the Written Opinion, amendment has been proposed to the description and claims to overcome a number of formality problems. Following the proposed amendments, it is submitted that the description and claims are now clear.

We trust that a clear International Preliminary Examination Report will issue on this application. If, however, the proposed amendments and submissions do not overcome the objections, the applicant would appreciate an opportunity to respond further.

Yours respectfully

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Encls

ELECTROPHORESIS MEMBRANE SUPPORT AND MANIFOLD UNIT

Technical Field

The present invention relates to a membrane support and manifold adapted to accept one or more membranes suitable for use in an electrophoresis apparatus.

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Background Art

Membrane-based electrophoresis is a new technology originally developed for the separation of macromolecules such as proteins, nucleotides and complex sugars. This unique preparative electrophoresis technology originally developed for macromolecule separation utilises tangential flow across polyacrylamide membranes with an electric field or potential applied across the membranes. The general design of the system facilitates the purification of proteins and other macromolecules under near native conditions. This results in higher yields and excellent purity. The process provides a high purity, scalable separation that is faster, cheaper and higher yielding than current methods of macromolecule separation. Furthermore, the technology offers the potential to concurrently purify and decontaminate macromolecule solutions.

To scale up any separation technology for commercial applications, new apparatus design and technology are often required. To scale up membrane-based electrophoresis based on the original Gradiflow™ technology developed by Gradipore Limited (see US 6413402; US 6328869; US 5039386; US 5650055 and WO 02/24314), the present inventors found it was necessary to significantly alter many of the components of the apparatus to accommodate large membranes and overcome problems encountered with scale-up.

The present inventors have developed an improved membrane support and manifold for use in larger scale membrane-based electrophoresis.

Disclosure of Invention

In a first aspect, the present invention provides an electrophoresis membrane support comprising:

30 a substantially planar member having four boundaries, an upper face and a lower face;
an inlet port disposed near one boundary;
an outlet port disposed opposite the inlet port near an opposite boundary;

(cathode). Conversely, a negatively-charged ion will move toward the positive electrode (anode).

Membrane-based electrophoresis apparatus (Gradiflow™) developed by, or in association with, Gradipore Limited, Australia are suitable for performing the methods 5 described herein and are fully disclosed in commonly assigned US 6413402, US 6328869, US 5039386, US 5650055 and WO 02/24314. The apparatus may include a cartridge which houses a number of membranes forming at least two chambers, a cathode and an anode in respective electrode chambers connected to a suitable power supply, reservoirs for samples, buffers and electrolytes, pumps for passing samples, 10 buffers and electrolytes, and cooling means to maintain samples, buffers and electrolytes at a required temperature during electrophoresis. The cartridge typically contains at least three substantially planar membranes disposed and spaced relative to each other to form two chambers through which sample or solvent can be passed. A separation membrane is disposed between two outer membranes (termed restriction membranes as 15 their molecular mass cut-offs are usually smaller than the cut-off of the separation membrane). When the cartridge is installed in the apparatus, the restriction membranes are typically located adjacent to an electrode. One example of a cartridge is described in AU 738361.

In the apparatus, ion-permeable barriers that substantially prevent convective 20 mixing between the adjacent chambers of the manifold according to the present invention are placed in an electric field and components of the sample are selectively transported through the ion-permeable barriers. The particular ion-permeable barriers used will vary for different applications and generally have characteristic average pore sizes and pore size distributions and/or isoelectric points allowing or substantially 25 preventing passage of different components.

Having outlined some of the principles of operation of an apparatus, a manifold and separation unit will be described.

Figure 1 shows a preferred concept of the present invention providing transverse flow of fluid in adjacent chambers formed by three membranes. The change 30 in flow configuration, although difficult to develop and engineer, results in significant improvement in electrophoresis separation. A transverse flow allows more efficient flow of material over the membranes, reduced void volumes, less gas build-up, and increased separation efficiency. A new membrane support to direct and control liquid flow had to be developed that would allow this new configuration of the electrophoresis separation 35 unit. It will be appreciated, however, that the membrane support can be used alone in an

Claims:

1. An electrophoresis membrane support comprising:

a substantially planar member having four boundaries, an upper face and a lower face;

5 an inlet port disposed near one boundary;

an outlet port disposed opposite the inlet port near an opposite boundary;

spacers positioned between the inlet port and outlet port adapted to support a membrane positioned on the upper face or on the lower face of the member;

interstitial space disposed between the spacers capable of allowing flow of fluid

10 therein;

inlet means in fluid communication with the inlet port and the interstitial space;

outlet means in fluid communication with the interstitial space and the inlet port, the inlet and outlet means adapted to allow flow of fluid along the interstitial space;

first flow port disposed near one boundary; and

15 second flow port disposed opposite the first flow port near an opposite boundary, the first and second flow ports direct flow of fluid to or from the electrophoresis apparatus.

20 2. The support according to claim 1 wherein two supports are adapted to be assembled in an electrophoresis apparatus or cartridge to form transverse fluid flow paths along respective interstitial spaces formed by each support.

3. The support according to claim 1 or 2 wherein at least some of the inlet, outlet and flow ports are provided as channels formed in each respective boundary of the member.

25 4. The support according to claim 1 or 2 wherein at least some of the inlet, outlet and flow ports are formed as a plurality of ports or holes in the member.

5. The support according to claim 1 or 2 wherein the inlet and outlet ports are formed as non-circular ports to assist in movement of fluid in the port to enter the inlet or outlet means.

30 6. The support according to any one of claims 1 to 5 wherein the spacers are formed as a plurality of substantially planar parallel members running from the inlet means to the outlet means.

7. The support according to any one of claims 1 to 6 wherein the inlet means is formed by a series of flow channels directing fluid from the inlet port to the interstitial space.
8. The support according to any one of claims 1 to 7 wherein the outlet means is formed by a series of flow channels directing fluid from the interstitial space to the outlet port.
- 5 9. The support according to any one of claims 1 to 8 being substantially square in shape with the ports disposed near each of the four boundaries forming the square.
10. The support according to any one of claims 1 to 9 further comprising one or more drain ports.
11. The support according to claim 10 wherein the drain ports are in communication with 10 drain channels adapted to receive fluid escaped from the ports or interstitial spaces.
12. An electrophoresis separation unit comprising:
 - a first manifold having at least one inlet port and one outlet port;
 - a second manifold having at least one inlet port and one outlet port;
 - a plurality of electrophoresis membrane supports according to any one of claims 1 to 11 disposed between the first manifold and the second manifold; and
 - a plurality of ion-permeable membranes disposed between the membrane supports forming a plurality of adjacent flow chambers between the membranes;
 - wherein in use the direction of flow of fluid in one flow chamber is transverse to the direction of flow of another flow chamber.
- 20 13. The separation unit according to claim 12 having two membrane supports and three membranes forming a first flow chamber and a second flow chamber.
14. The separation unit according to claim 13 wherein the first manifold includes a first inlet port and first outlet port in fluid communication with the first chamber and a second inlet port and second outlet port in communication with the second chamber.
- 25 15. The separation unit according to claim 13 wherein the second manifold includes a first inlet port and first outlet port in communication with the first chamber and a second inlet port and second outlet port in communication with the second chamber.
16. The separation unit according to any one of claims 12 to 15 further containing an electrode associated with both the first and second manifolds.

17. The separation unit according to claim 12 comprising:

a first manifold having a first inlet port and first outlet port, a second inlet port and second outlet port, and a third inlet port and a third outlet port;

5 a second manifold having an inlet port and outlet port;

a first ion-permeable membrane disposed adjacent the first manifold;

10 a second ion-permeable membrane disposed adjacent the first ion-permeable membrane;

a third ion-permeable membrane disposed adjacent the second manifold;

15 a first membrane support disposed between the first ion-permeable membrane and the second ion-permeable membrane;

a second membrane support disposed between the third ion-permeable membrane and the second ion-permeable membrane;

a first fluid chamber adapted to receive fluid in a first stream disposed between the first ion-permeable membrane and the second ion-permeable membrane;

20 a second fluid chamber adapted to receive fluid in a second stream between the second ion-permeable membrane and the third ion-permeable membrane;

a first electrolyte chamber containing a first electrode disposed between the first ion-permeable membrane and the first manifold; and

25 a second electrolyte chamber containing a second electrode disposed between the third ion permeable membrane and the second manifold;

wherein the first inlet port and first outlet port are in fluid communication with the first fluid chamber, the second inlet port and second outlet port are in fluid communication with the second fluid chamber, the third inlet port and third outlet port are in fluid communication with the first electrolyte chamber, and the inlet port and outlet port are in fluid communication with the second electrolyte chamber, and

wherein in use the direction of flow of the first steam is transverse to the direction of flow of the second stream.

18. The separation unit according to any one of claims 12 to 17 wherein at least one ion permeable membrane is a hydrogel membrane, an endo-electro-osmosis

30 membranes capable of controlling the bulk movement of water, an isoelectric membrane, or a membrane having defined pore size or pore size distribution.